UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
10/526,425	03/03/2005	Tsuneko Okazaki	80161(302730)	9673

21874 7590 01/22/2008
EDWARDS ANGELL PALMER & DODGE LLP
P.O. BOX 55874
BOSTON, MA 02205

EXAMINER	
HILL, KEVIN KAI	

ART UNIT	PAPER NUMBER
1633	

MAIL DATE	DELIVERY MODE
01/22/2008	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/526,425	OKAZAKI ET AL.	
	Examiner	Art Unit	
	Kevin K. Hill, Ph.D.	1633	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *01 October 2007*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-7,13,14,17,18,23-29,31-45,50,52,54 and 56* is/are pending in the application.

4a) Of the above claim(s) *2,17,18,23-29,31-45,50,52,54 and 56* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1,3-7,13 and 14* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☒ All b)☐ Some * c)☐ None of:

1.☒ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

Detailed Action

Applicant's response to the Requirement for Restriction, filed on October 1, 2007 is acknowledged.

Applicant has elected the invention of Group I, claim(s) 1, 3-7 and 13-14, drawn to a method of producing a circular mammalian artificial chromosome.

Within Group I, Applicant has elected the insertion sequence species "lox P site", as recited in Claim 13.

Election of Applicant's invention(s) was made with traverse.

Applicant argues that:

a) the examiner has failed to apply correctly PCT Rule 13.2, in that both Groups I and II are drawn to production methods of mammalian artificial chromosomes and encompass three steps that are essentially the same; and

b) there would be no burden to search and examine all the claims in a single application.

Applicants' arguments have been fully considered but are not found persuasive.

With respect to a), the Examiner has explained in the Requirement for Restriction that the special technical feature of Groups II and IV is a linear artificial chromosome comprising a telomere not present in the circular artificial chromosome. The Group VI method steps recited special technical features to make a micro-cell not present in the Group V method. Furthermore, Applicant is already aware that the process for constructing an artificial mammalian chromosome which involves the step of transferring a first vector containing a mammalian centromere sequence and a second vector containing a functional sequence into a mammalian host cell had already been known in the art, as discussed in the International Search Report of PCT/JP03/11134.

With respect to b), MPEP §803 states that "If the search and examination of all the claims in an application can be made without serious burden, the examiner must examine them on the merits, even though they include claims to independent or distinct inventions."

In the instant case a serious burden exists since each limitation, directed to micro-cells, linear artificial chromosomes comprising telomeres, circular artificial chromosomes, and transgenic animals requires a separate, divergent, and non co-extensive search and examination of the patent and non-patent literature. For instance, a search and consideration of the prior art as it relates to circular artificial chromosomes would not be adequate to uncover prior art related to non-integrative transgenic animals.

Further, a search and examination of all the claims directed to all embodiments involves different considerations of novelty, obviousness, written description, and enablement for each claim. In view of these requirements, it is the Examiner's position that searching and examining all of the claims including limitations to micro-cells, linear artificial chromosomes comprising telomeres, circular artificial chromosomes, and transgenic animals in the same application presents a serious burden on the Examiner for the reasons given above and in the previous Restriction Requirement.

The requirement is still deemed proper and is therefore made FINAL.

Claims 2, 17-18, 23-29, 31-45, 50, 52, 54 and 56 are pending but withdrawn from further consideration pursuant to 37 CFR 1.142(b) as being drawn to a non-elected invention, there being no allowable generic or linking claim.

Claims 1, 3-7 and 13-14 are under consideration.

Priority

This application is a 371 of PCT/JP03/11134, filed September 1, 2003. Acknowledgment is made of Applicant's claim for foreign priority under 35 U.S.C. 119(a)-(d). Certified copies of the foreign patent applications Japan 2002-258114, filed September 3, 2002 and Japan 2002, filed November 22, 2002 are filed with the instant application. Certified English translations of said foreign applications have not been provided.

Information Disclosure Statement

Applicant has filed Information Disclosure Statements on March 3, 2005, January 9, 2006 and August 6, 2007.

The information disclosure statement filed March 3, 2005 fails to comply with 37 CFR 1.98(a)(3) because it does not include a concise explanation of the relevance, as it is presently understood by the individual designated in 37 CFR 1.56(c) most knowledgeable about the content of the information, of each patent listed that is not in the English language. In particular, English translations of citations FC, GA and HB have not been provided. They have been placed in the application file, but the information referred to therein has not been considered.

The Examiner was able to consider the remaining citations to the extent of time allowable. The signed and initialed PTO Forms 1449 are mailed with this action.

Claim Objections

1. **Claim 3 are objected to because of the following informalities**: the claim is drawn to non-elected subject matter, specifically claim 2. It would be remedial to draft the claims in independent form. Appropriate correction is required.

Claim Rejections - 35 USC § 112

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

2. **Claims 1, 3-7 and 13-14 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement.** The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

The claimed invention is directed to a method of making a mammalian artificial chromosome, wherein the artificial chromosome comprises an insulator sequence. At issue for the purpose of written description requirements is the lack of adequate description of an insulator sequence.

> *Vas-cath Inc. v. Mahurkar*, 19USPQ2d 1111, clearly states that Applicant must convey with reasonable clarity to those skilled in the art that, as of the filing date sought, he or she was in possession of the invention. The invention is, for purposes of the 'written description' inquiry, whatever is now claimed." (See page 1117.) The specification should "clearly allow persons of ordinary skill in the art to recognize that (he or she) invented what is claimed." (See *Vas-cath* at page 1116).

To provide adequate written description and evidence of possession of a claimed genus, the specification must provide sufficient distinguishing identifying characteristics of the genus. The factors to be considered include disclosure of compete or partial structure, physical and/or chemical properties, functional characteristics, structure/function correlation, methods of making the claimed product, or any combination thereof.

In analyzing whether the written description requirement is met for genus claims, it is first determined whether a representative number of species have been described by their complete structure.

> The disclosure of a single species is rarely, if ever, sufficient to describe a broad genus, particularly when the specification fails to describe the features of that genus, even in passing. (see *In re Shokal* 113USPQ283(CCPA1957); *Purdue Pharma L.P. vs Faulding Inc.* 56 USPQ2nd 1481 (CAFC 2000).

In the instant case, the specification discloses the use of the human β globin locus control region (LCR) comprising HS1-HS5 (pg 52, Example 15).

Next, then, it is determined whether a representative number of species have been sufficiently described by other relevant identifying characteristics (i.e. other than nucleotide

sequence), specific features and functional attributes that would distinguish different members of the claimed genus. In the instant case, the only other identifying characteristic is that "the insulator sequence is a base sequence characterized by exhibiting an enhancer blocking effect (expressions of neighboring genes are not affected by each other) or a chromosome boundary effect (a region assuring the gene expression and a region suppressing the gene expression are separated with each other). Usable insulator sequences are not particularly limited. It is possible to use not only an insulator, which has been identified as an insulator, but also a sequence obtained by providing modification for the sequence as long as the expected effect (the increase in promoting the expression of target gene or the increase in the gene introduction efficiency) is not reduced. (pg 22, lines 16-34).

The Revised Interim Guidelines state:

"The claimed invention as a whole may not be adequately described if the claims require an essential or critical element which is not adequately described in the specification and which is not conventional in the art" (col. 3, page 71434), "when there is substantial variation within the genus, one must describe a sufficient variety of species to reflect the variation within the genus", "in an unpredictable art, adequate written description of a genus which embraces widely variant species cannot be achieved by disclosing only one species within the genus" (col. 2, page 71436).

An applicant shows possession of the claimed invention by describing the claimed invention with all of its limitations using such descriptive means as words, structures, figures, diagrams, and formulas that fully set forth the claimed invention. *Lockwood v. American Airlines, Inc.*, 107 F.3d 1565, 1572, 41 USPQ2d 1961, 1966 (Fed. Cir. 1997).

Possession may also be shown in a variety of ways including description of an actual reduction to practice, or by showing that the invention was "ready for patenting" such as by the disclosure of drawings or structural chemical formulas that show that the invention was complete, or by describing distinguishing identifying characteristics sufficient to show that the applicant was in possession of the claimed invention. See, e.g., *Pfaff v.*

Wells Elecs., Inc., 525 U.S. 55, 68, 1 19 S.Ct. 304, 312, 48 USPQ2d 1641, 1647 (1998), *Regents of the University of California v. Eli Lilly*, 119 F.3d 1559, 1568, 43 USPQ2d 1398, 1406 (Fed. Cir. 1997)*, *Amgen, Inc. v. Chugai Pharmaceutical*, 927 F.2d 1200, 1206, 18 USPQ2d 1016, 1021 (Fed. Cir. 1991) (one must define a compound by "whatever characteristics sufficiently distinguish it").

Therefore, conception is not achieved until reduction to practice has occurred, regardless of the complexity or simplicity of the method of isolation. See *Fiers v. Revel*, 25 USPQ2d 1602 at 1606 (CAFC 1993) and *Amgen Inc. v. Chugai Pharmaceutical Co. Ltd.*, 18 USPQ2d 1016. One cannot describe what one has not conceived. See *Fiddes v. Baird*, 30 USPQ2d 1481, 1483. *In Fiddes*, claims directed to mammalian FGF's were found to be unpatentable due to lack of written description for that broad class. The specification provided only the bovine sequence.

Without a correlation between structure and function, the claim does little more than define the claimed invention by function. That is not sufficient to satisfy the written description requirement. *See Eli Lilly*, 119 F.3d at 1568, 43 USPQ2d at 1406 ("definition by function ... does not suffice to define the genus because it is only an indication of what the gene does, rather than what it is").

The specification does not disclose any identifying structural characteristic as to how an artisan would *a priori* know that a given nucleic acid sequence is, or comprises, an insulator sequence. At the time of the invention, insulator elements were a novel class of regulatory elements only known by functional definitions (Bell et al, Curr. Op. Genetics & Dev. 9:191-198, 1999). The first is position-dependent enhancer-blocking activity: insulators block enhancer action only when placed between an enhancer and promoter but not upstream or downstream of an enhancer-promoter pair. The second insulator activity is an ability to form chromatin boundaries and therefore confer position-independent transcription to transgenes stably integrated in the genome. Applicant presently introduces a third functional definition: the

increase in the gene introduction efficiency. However, insulators were "still something of a black box" (pg 196, col. 2, ¶3), and no specific nucleotide sequence or structure was known as being shared with all insulator sequences so as to establish a "hallmark signature" or canonical sequence. Thus, each insulator sequence will possess a distinctly different structure, and vary greatly in structure with another, yet-to-be-established insulator.

Based on applicant's specification, the skilled artisan cannot envision the detailed chemical structure of the nucleotide sequences which encode an insulator sequence as defined by the specification or encompassed by the claims. The one insulator sequence species specifically disclosed, the human β globin locus control region (LCR) comprising HS1-HS5, is not representative of the genus because the genus is highly variant.

Accordingly, given that the specification does not teach what is the complete structure of the exceptionally broadly-defined "insulator sequence" genus, this limited information is not deemed sufficient to reasonably convey to one skilled in the art that the applicant is in possession of the required starting materials, that is the broad genus of insulator sequences, to perform the necessary active steps and effect the claimed method, at the time the application was filed.

Thus, for the reasons outlined above, it is concluded that the claims do not meet the requirements for written description under 35 U.S.C. 112, first paragraph. Applicant is reminded that *Vas-Cath* makes clear that the written description provision of 35 U.S.C. §112 is severable from its enablement provision (see page 1115).

3. **Claims 1, 3-7 and 13-14 are rejected under 35 U.S.C. 112, first paragraph**, because the specification, while being enabling for a method of making a mammalian artificial chromosome, the method comprising:

i) a first step of introducing a first vector being circular in form comprising a mammalian centromere sequence and a second vector being circular in form comprising an insertion sequence for specifically inserting a sequence of interest and a β-globin locus control region insulator sequence into a mammalian host cell;

ii) a second step of selecting transformed cells; and

iii) a third step of selecting a cell containing a mammalian artificial chromosome from the selected transformed cells,

does not reasonably provide enablement for a genus of insulator sequences. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to practice the invention commensurate in scope with these claims.

While determining whether a specification is enabling, one considers whether the claimed invention provides sufficient guidance to make and use the claimed invention. If not, whether an artisan would have required undue experimentation to make and use the claimed invention and whether working examples have been provided. When determining whether a specification meets the enablement requirements, some of the factors that need to be analyzed are: the breadth of the claims, the nature of the invention, the state of the prior art, the level of one of ordinary skill, the level of predictability in the art, the amount of direction provided by the inventor, the existence of working examples, and whether the quantity of any necessary experimentation to make or use the invention based on the content of the disclosure is "undue" (*In re Wands*, 858 F.2d 731, 737, 8 USPQ2ds 1400, 1404 (Fed. Cir. 1988)). Furthermore, USPTO does not have laboratory facilities to test if an invention will function as claimed when working examples are not disclosed in the specification. Therefore, enablement issues are raised and discussed based on the state of knowledge pertinent to an art at the time of the invention. And thus, skepticism raised in the enablement rejections are those raised in the art by artisans of expertise.

The Breadth of the Claims and The Nature of the Invention

The breadth of the claim is large for encompassing a genus of nucleic acid structures identified only by their functional properties, specifically insulator elements. The inventive concept in the instant application is the use of a human β-globin insulator element in a method of making a mammalian artificial chromosome.

The Existence of Working Examples and The Amount of Direction Provided by the Inventor

The specification discloses that "the insulator sequence is a base sequence characterized by exhibiting an enhancer blocking effect (expressions of neighboring genes are not affected by

each other) or a chromosome boundary effect (a region assuring the gene expression and a region suppressing the gene expression are separated with each other). Usable insulator sequences are not particularly limited. It is possible to use not only an insulator, which has been identified as an insulator, but also a sequence obtained by providing modification for the sequence as long as the expected effect (the increase in promoting the expression of target gene or the increase in the gene introduction efficiency) is not reduced. (pg 22, lines 16-34).

The State of the Prior Art, The Level of One of Ordinary Skill and The Level of Predictability in the Art

At the time of the invention, insulator elements were a novel class of regulatory elements only known by functional definitions (Bell et al, Curr. Op. Genetics & Dev. 9:191-198, 1999). The first is position-dependent enhancer-blocking activity: insulators block enhancer action only when placed between an enhancer and promoter but not upstream or downstream of an enhancer-promoter pair. The second insulator activity is an ability to form chromatin boundaries and therefore confer position-independent transcription to transgenes stably integrated in the genome. However, insulators were "still something of a black box" (pg 196, col. 2, ¶3), and no specific nucleotide sequence or structure was known as being shared with all insulator sequences so as to establish a "hallmark signature" or canonical sequence. While Applicant further defines an insulator element as having the functional property of increasing the yield of product mammalian artificial chromosomes in an *in vivo* recombination cloning reaction, the prior art does not teach this functional property of insulator elements. Thus, each insulator sequence will possess a distinctly different structure, and vary greatly in structure with another, yet-to-be-established insulator.

The mechanism(s) by which insulator elements function were not well understood, and conflicting models existed to account for their properties (Zhan et al, Human Genetics 109:471-478, 2001; pg 473, col. 2, Models). The art recognized that insulator elements were complex and that the mechanism of boundary action remains obscure (pg 475, col. 2, Complexity). Furthermore, the concept of insulators has been subject to change as their functional definitions change upon continued research of the phenomena. Many questions still remain to be answered

before we understand the structure and role of chromatin insulators, but their unique properties lend themselves to endless possibilities for controlling eukaryotic gene expression. Insulators might also come in different "flavors" and "strengths", giving rise to a hierarchy of chromatin domains with variable abilities to affect enhancer-promoter interactions. The characterization of these elements and definition of their roles in the regulation of gene expression and chromatin structure will be an interesting challenge (pg 476, col. 1).

Thus, one of ordinary skill in the art would recognize considerable unpredictability in knowing a priori if a given nucleic acid sequence is or is not an insulator element because the functional definitions of insulator elements are not well-established, and instead has "changed dramatically". No one has been able to elucidate the whole picture yet (Zhan et al, pg 476, col. 1, Conclusions).

The Quantity of Any Necessary Experimentation to Make or Use the Invention

Thus, the quantity of necessary experimentation to make or use the invention as claimed, based upon what is known in the art and what has been disclosed in the specification, will create an undue burden for a person of ordinary skill in the art to demonstrate that a given nucleic acid sequence comprises the functional properties of an insulator because the art did not have a unifying understanding or functional definition of an insulator sequence, did not possess a consensus nucleic acid "hallmark" sequence that would identify a nucleic acid as possessing insulator properties, and did not teach that insulator sequences might increase the gene introduction efficiency when cloning a desired gene into a mammalian artificial chromosome. In the absence of a specific disclosure that would clearly identify an insulator sequence in a population of nucleic acids, the artisan would essentially have to perform the claimed method just to ascertain whether or not a given nucleic acid sequence would possess a functional property that would clearly identify the sequence as an insulator.

In conclusion, the specification fails to provide any guidance as to how an artisan would have dealt with the art-recognized limitations of the claimed method commensurate with the scope of the claimed invention and therefore, limiting the claimed invention to a method of making a mammalian artificial chromosome, the method comprising:

i) a first step of introducing a first vector being circular in form and comprising a mammalian centromere sequence, and a second vector being circular in form and comprising an insertion sequence for specifically inserting a sequence of interest and an β-globin locus control region insulator sequence into a mammalian host cell;

ii) a second step of selecting transformed cells; and

iii) a third step of selecting a cell containing a mammalian artificial chromosome from the selected transformed cells, is proper.

Claim Rejections - 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

4. **Claims 1, 3-7 and 13-14 are rejected under 35 U.S.C. 112, second paragraph**, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

With respect to claim 1 (and dependent claims), the claims are vague in that no step(s) in the claimed method refers back to or recapitulates the preamble of the claim. Applicants recite a method of making a mammalian artificial chromosome, but no step is recited that actually accomplishes the preamble. It is unclear if additional, undisclosed steps are a part of the claimed method and therefore the metes and bounds of the claimed subject matter are unclear.

With respect to claim 1, the grammatical structure of the first step is unclear because a plurality of elements, delimited only by the conjunction "and", are to be introduced into a mammalian host cell. The claim does not clearly identify which elements belong to the respective vector(s).

With respect to claim 1 (and dependent claims), the claims are incomplete for omitting essential steps, such omission amounting to a gap between the steps. See MPEP § 2172.01. The omitted steps are: the means by which the cell containing the mammalian artificial chromosome is to be selected from the transformed cells.

With respect to claim 3, the recites the limitation "the selection marker gene" in reference to the selection step of claim 1. There is insufficient antecedent basis for this limitation in the claim. The specification discloses that the second vector may also comprise a selection marker gene (pg 16, lines 33-34), and thus it is unclear if "the selection method step is carried out by using the selection marker gene" of the first vector or the second vector.

With respect to claims 5-6, the claims recite "a sequence derived from a human chromosome alpha satellite region"..."of a human chromosome 21". The claims are vague and indefinite in that the metes and bounds of the term "derived from" are unclear. It is unclear the nature and number of steps required to obtained a "derivative" of the alpha satellite region. The term implies a number of different steps that may or may not result in a change in the functional characteristics of the nucleic acid(s) from the source that it is "derived from". It would be remedial to amend the claim language to use the term "obtained from", which implies a more direct method of acquiring the nucleic acid.

Additionally, how would the artisan know that a given nucleic acid sequence, e.g. a nucleotide, a di-nucleotide, or an oligonucleotide, was "derived from" a human chromosome alpha satellite region? The specification discloses that the sequence derived from a human chromosome alpha satellite region may be further modified by substitution, deletion, insertion and/or addition of one or a plurality of bases in the sequence of interest (pg 17, lines 26-30). With such latitude in nucleic acid sequence permutations, essentially any nucleic acid sequence would fulfill the instant limitation. The examiner also notes that, as presently stated, a single nucleotide, e.g. A, C, T or G, or a repeated sequence thereof, is sufficient to meet the claim as the nucleotide could be reasonably "derived from a human chromosome alpha satellite region"..."of a human chromosome 21".

With respect to claims 1 and 13, the claims are incomplete for omitting essential elements, such omission amounting to a gap between the elements. See MPEP § 2172.01. The omitted elements are: a loxP site present in the first vector. Recombination cloning requires two compatible recombination sites, wherein a first recombination site is present in the donor nucleic acid and a second recombination site is present in the target nucleic acid. The instantly claimed method does not recite the first vector to comprise a loxP site necessary for recombination

cloning, nor the do the claims recite an alternative means by which the donor nucleic acid is to be inserted into the lox P "insertion sequence" of the second vector. Furthermore, the claims do not recite the presence of Cre recombinase that acts upon loxP sites to mediate recombination, nor how such Cre will be introduced into the mammalian host cell.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

The factual inquiries set forth in *Graham* **v.** *John Deere Co.*, 383 U.S. 1, 148 USPQ 459 (1966), that are applied for establishing a background for determining obviousness under 35 U.S.C. 103(a) are summarized as follows:

1. Determining the scope and contents of the prior art.
2. Ascertaining the differences between the prior art and the claims at issue.
3. Resolving the level of ordinary skill in the pertinent art.
4. Considering objective evidence present in the application indicating obviousness or nonobviousness.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

5. **Claims 1, 3-6 and 13 are rejected under 35 U.S.C. 103(a)** as being unpatentable over Mejia et al (Genomics 70(2):165-170, 2000; *of record in IDS, AE), in further view of Waye et al (Mol. and Cell. Biol. 6(9):3156-3165, 1986), Ikeno et al (Human Mol. Gen. 3(8):1245-1257, 1994; *of record in IDS, CE) and Perkins et al (US 2003/0119104 A1).

Determining the scope and contents of the prior art.

Mejia et al teach a method of making mammalian artificial chromosomes, the method comprising a step of introducing into a prokaryotic host cell a first vector being circular in form and comprising a mammalian centromere sequence, and a second vector being in circular form and comprising an insertion sequence for specifically inserting a sequence of interest. The method further comprises a step of selecting the transformed cells and selecting a cell containing a mammalian artificial chromosome from the selected transformed cells (pg 167, Figure 2). The first vector comprises a selection marker gene, specifically chloramphenicol-resistance (pg 167, Figures 1 and 2), wherein the selection step for the transformed cells is carried out by using the chloramphenicol-resistance marker gene. The mammalian centromere sequence comprises 220kb of α satellite DNA from the human chromosome 17 centromere (pg 165, col. 1, ¶1). The insertion sequence is a loxP recombination site (pg 166, col. 1, ¶2).

Mejia et al do not teach the method being performed in mammalian cells, nor that the vector comprises an insulator sequence. However, at the time of the invention, Perkins et al disclosed a method of producing artificial chromosomes in eukaryotic cells (pgs 6-7, [0074]; pg 14, [0155]-pg 15, [0168]) comprising the use of recombinase enzymes (pg 4, [0051-56]; pg 13, [0145-149]), where the desired nucleic acids are co-transfected into a eukaryotic host cell (pg 14, [0161]). Insulator sequences may be introduced into the artificial chromosome to assist in the expression of the desired transgene or genomic locus, e.g. the β-globin HS4 insulator element (pg 17, [0192]-pg 18, [0196]). Eukaryotic host cells containing the artificial chromosome were selected via assays such as fluorescent in situ hybridization (FISH) (pg 28, Example 2, [0320]).

Neither Mejia et al nor Perkins et al teach the α satellite DNA from the human chromosome 17 centromere to comprise the sequence of SEQ ID NO:1, nor wherein the

centromere sequence comprises an 11-mer repeat unit derived from human chromosome 21. However, at the time of the invention, α satellite (alphoid) DNA was known in the prior art to form a functional centromere in a human artificial chromosome, wherein the presence of a centromere protein B sequence (CENP-B box) in the alphoid DNA is a requirement for the functional centromere. Waye et al taught the sequence of the human chromosome 17 centromere (pg 3159, Figure 3) comprising nucleic acid sequences 100% identical to SEQ ID NO:1. Furthermore, Ikeno et al taught a consensus CENP-B box nucleotide sequence from human chromosome 21 alphoid repeats (pg 1250, Table 1), wherein the human chromosome 17 centromere comprises an 11-mer repeat with 100% identity to the consensus sequence set forth by Ikeno et al (Waye et al; pg 3159, Figure 3, e.g., the first 20 nucleotides of monomer 10). Thus, while the human chromosome 17 centromere comprising an 11-mer repeat with 100% identity to the consensus sequence set forth by Ikeno et al of the consensus CENP-B box nucleotide sequence from human chromosome 21 alphoid repeats is an endogenous sequence, is not "derived from human chromosome 21" as claimed, the 11-mer repeat fulfills the structural identity of the claim, and thus necessarily fulfills the functional requirement of the centromeric feature of the invention. Neither the claims nor the specification disclose an essential feature of the 11-mer repeat that is present when "derived from human chromosome 21" that would not be present in the context of human chromosome 17.

Resolving the level of ordinary skill in the pertinent art.

People of the ordinary skill in the art will be highly educated individuals, possessing advanced degrees, including M.D.'s and Ph.D.'s. They will be medical doctors, scientists, or engineers. Thus, these people most likely will be knowledgeable and well-read in the relevant literature and have the practical experience in molecular biology, recombination cloning, making artificial chromosomes, creating transgenic cells. Therefore, the level of ordinary skill in this art is high.

Considering objective evidence present in the application indicating obviousness or nonobviousness.

It would have been obvious to one of ordinary skill in the art to substitute the prokaryotic cells as taught by Mejia et al with mammalian cells as taught by Perkins et al with a reasonable chance of success because the simple substitution of one known element for another would have yielded predictable results to one of ordinary skill in the art at the time of the invention. At the time of the invention, the ordinary artisan was well-aware that recombination cloning could be practiced in mammalian cells, and had the means to identify and select the mammalian cell containing the desired artificial chromosome. An artisan would be motivated to substitute the prokaryotic cells for mammalian cells because protocols for isolating large mammalian DNA chromosomes *in vitro* or from an *E. coli* host cell lysate is cumbersome, subject to significant DNA degradation, and results in quantitatively poor yields; whereas, *in vivo* recombination is efficient and obviates the need to purify recombination products prior to transduction into the mammalian host cell.

It also would have been obvious to one of ordinary skill in the art to include an insulator element in the mammalian artificial chromosome with a reasonable chance of success because all the claimed elements were known in the prior art and one skilled in the art could have combined the elements as claimed by known methods with no change in their respective functions, and the combination would have yielded predictable results to one of ordinary skill in the art at the time of the invention. An artisan would be motivated to combine an insulator element with artificial chromosomes because these sequences help to define boundaries in chromatin structure and thus minimize influence of chromatin position effect variegation and gene silencing on the expression of the target gene.

Thus, the invention as a whole is *prima facie* obvious.

6. **Claims 1 and 7 are rejected under 35 U.S.C. 103(a)** as being unpatentable over Mejia et al (Genomics 70(2):165-170, 2000; *of record in IDS, AE), in further view of Waye et al (Mol. and Cell. Biol. 6(9):3156-3165, 1986), Ikeno et al (Human Mol. Gen. 3(8):1245-1257, 1994; *of record in IDS, CE) and Perkins et al (US 2003/0119104 A1), as applied to claims 1, 3-6 and 13 above, and in further view of Bokkelen et al (U.S. Patent No. 5,695,967).

Determining the scope and contents of the prior art.

The prior cited art does not teach the mammalian centromere sequence to be about 50kb or less. However, at the time of the invention, Bokkelen et al disclosed a means of cloning iterations of mammalian centromeric alphoid DNA 2.7kb higher order repeats, e.g. 2.7kb, 5.4kb, 11kb, 22kb, 43kb, 86kb, 130kb and 174kb iterations (Figure 1; col. 6, lines 22-25).

Resolving the level of ordinary skill in the pertinent art.

People of the ordinary skill in the art will be highly educated individuals, possessing advanced degrees, including M.D.'s and Ph.D.'s. They will be medical doctors, scientists, or engineers. Thus, these people most likely will be knowledgeable and well-read in the relevant literature and have the practical experience in molecular biology, recombination cloning, making artificial chromosomes, creating transgenic cells. Therefore, the level of ordinary skill in this art is high.

Considering objective evidence present in the application indicating obviousness or nonobviousness.

It would have been obvious to substitute the mammalian centromere sequence length of Mejia et al with a mammalian centromere sequence length of about 50kb or less as taught be Bokkelen et al with a reasonable chance of success because the simple substitution of one known element for another would have yielded predictable results to one of ordinary skill in the art at the time of the invention. At the time of the invention, the ordinary artisan knew how to design specific lengths, composition, orientation and phasing of centromeric 171 base-pair alphoid DNA monomers to create arrays of specific lengths, e.g. 171 base-pairs to about 270kb. An artisan would be motivated to make a mammalian centromere sequence of about 50kb or less because alphoid DNA of larger sizes are difficult to clone and stably propagate because of the tendency of tandemly repetitive DNA to recombine into smaller arrays. Furthermore, the smaller length would provide more room in the artificial chromosome for the artisan to incorporate larger genes and/or regulatory elements.

Thus, the invention as a whole is *prima facie* obvious.

7. **Claims 1 and 14 are rejected under 35 U.S.C. 103(a)** as being unpatentable over Mejia et al (Genomics 70(2):165-170, 2000; *of record in IDS, AE), in further view of Waye et al (Mol. and Cell. Biol. 6(9):3156-3165, 1986), Ikeno et al (Human Mol. Gen. 3(8):1245-1257, 1994; *of record in IDS, CE), Perkins et al (US 2003/0119104 A1) and Bokkelen et al (U.S. Patent No. 5,695,967), as applied to claims 1, 3-7 and 13 above, and in further view of Cooke et al (WO 00/18941).

Determining the scope and contents of the prior art.

The prior cited art does not teach the ratio of the first vector to the second vector to be in the range from about 10:1 to about 1:10 molecular ratio. However, at the time of the invention, Cooke et al disclosed a method of making mammalian artificial chromosomes, wherein the first and second nucleic acids may be mixed extra-cellularly before co-introduction into a competent host cells wherein recombination cloning takes place, wherein the cells may be mammalian cells (pg 15, lines 1-20; pg 16, lines 15-20). Cooke et al disclosed formulating 1.3:1 ratio (pg 25, lines18-19), as well as a 10:1 ratio (pg 29, line 20), of first vector to second vector.

Ascertaining the differences between the prior art and the claims at issue.

Cooke et al do not disclose the molecular ratio to be about 1:10; however, absent evidence to the contrary, nothing non-obvious is seen with this ratio because it is routine for the artisan to optimize relative ratios between the components of a cloning reaction so as to improve the yield of the desired reaction product.

Resolving the level of ordinary skill in the pertinent art.

People of the ordinary skill in the art will be highly educated individuals, possessing advanced degrees, including M.D.'s and Ph.D.'s. They will be medical doctors, scientists, or engineers. Thus, these people most likely will be knowledgeable and well-read in the relevant literature and have the practical experience in molecular biology, recombination cloning, making artificial chromosomes, creating transgenic cells. Therefore, the level of ordinary skill in this art is high.

Considering objective evidence present in the application indicating obviousness or nonobviousness.

It would have been obvious to one of ordinary skill in the art to try adjusting the molecular ratio of the first and second vector in a cloning reaction to be in the range from about 10:1 to about 1:10 molecular ratio because "a person of ordinary skill has good reason to pursue the known options within his or her technical grasp. If this leads to the anticipate success, it is likely that product not of innovation but of ordinary skill and common sense." Adjusting the relative ratios between a first (donor) nucleic acid and a second (target) nucleic acid in a molecular cloning reaction has long been practiced in the art. Furthermore, Cooke et al disclose working examples wherein the first and second vectors are in molecular ratios between 1.3:1 and 10:1. An artisan would be motivated to formulate the molecular ratio of the first and second vector in a cloning reaction to be in the range from about 10:1 to about 1:10 so as to optimize the yield of the desired product artificial chromosome in the transformed cell.

Thus, the invention as a whole is *prima facie* obvious.

Conclusion

8. No claims are allowed.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Kevin K. Hill, Ph.D. whose telephone number is 571-272-8036. The examiner can normally be reached on Monday through Friday, between 9:00am-6:00pm EST.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Joseph T. Woitach can be reached on 571-272-0739. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

Q. JANICE LI, M.D.
PRIMARY EXAMINER